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Comparison of wild and farmed sea bass (*Dicentrarchus labrax* L) lipid quality

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Abstract

The aim of the present work was to compare the fatty acids profiles of wild and farmed Mediterranean sea bass (*Dicentrarhus labrax* L.). Wild fish exhibited higher moisture content 74.6 (±1.1%) compared to the farmed (69.1 (±1.8%) Total fillet lipid content was and (1.68% ±1.9) and (7.31% ±1.59) in the wild and farmed fish, respectively. Total perivisceral fat was higher in farmed (25.87 ±5.78) compared to wild sea bass (0.11 ±0.02). Biochemical differences were also observed in the fatty acid profile of fillet and perivisceral fat. Docosahexaenoic acid (DHA, C22:6 n-3) was dominant in wild fish fillets instead of linoleic acid (C18:2 n-6) in the farmed fish. An increased presence of linoleic acid and a reduction of n-3/n-6 ratio in the fillets of farmed sea bass, most likely resulted from fish feeds rich in terrestrial plant oils, which compromised the nutritional quality of farmed fish. In the perivisceral fat, the dominant fatty acid was C18:1n-9 in both wild and farmed fish, followed by C16:1n-7, C22:1n-9 and C20:1n-9 in wild, and C20:1n-9, C22:1n-9 and C16:1n-7 in farmed fish. The results of the present work indicate that compared to the farmed fish, the wild sea bas exhibits significantly higher concentration of fatty acids with well known health benefits.

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Keywords: Fish oil; fatty acids; DHA; metabolism; Dicentrarchus labrax

1. Introduction

During the last twenty years, aquaculture sector has contributed to human nutrition providing fish of high nutritional quality in low prices. Moreover, farmed fish satisfied the increasing demand for fish proteins and indirectly protected natural stocks from further exploitation. This increasing demand and the dwindling of wild fish stocks require further development of the aquaculture sector in order to sustain the

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supply of nutritional products rich in fatty acids, amino-acids, trace elements and other valuable nutritional substances for the human diet.

The nutritional value of fish is attributed to the amino-acids, trace elements, vitamins, but most importantly, to their fatty acids (FAs) [1-3]. Recently, a reduction in the utilisation of fish oil in the manufacturing of aquaculture feeds is reflected in the FAs of farmed fish. The aim of the present work was to compare the fatty acids profile of wild and farmed sea bass flesh.

2. Materials and Methods

Forty sea bass specimens were examined originating from the delta of River Kalamas (NW Greece). The age of the fish was 1+ to 2+. Twenty of them were on-grown in an intensive fish farm in Valtos Bay (age of 22 months) and the other twenty were fished in the nearby lagoon of Loutsa-Papadia, which is situated in the coastal area of the Ionian Sea.

Fish feeds supplied to the farmed bass consisted (according to the label) of 43% proteins, 20% lipids, 16% carbohydrates, 7.4% ash, 1.6% fibres, 10% moisture, vitamins and trace elements. Raw materials were fish meal, fish oil, wheat flour, corn gluten, vitamins and minerals.

Initially, forty farmed and eighty wild specimens were collected in May 2008 using seine net and steady traps. They were killed with thermal shock in 0 ^{o}C and immediately transported in ice to the lab. Subsequently, fish were cleaned with fresh water and twenty farmed specimens were randomly selected. Body weight and total length were measured to the closest mg and mm, respectively, before scaling, skinning, de-heading, eviscerating, removing of the perivisceral fat and filleting the edible part of the flesh. All body parts of each fish were weighted, marked, vacuum packed in plastic bags and stored in -30 ^{0}C before examined.

Body weight and total length were also recorded in all eighty wild bass specimens and subsequently they were de-headed, marked and otoliths were removed. Otoliths were placed dry in plastic bags and age was determined within 48 h. Throughout this time period, all body parts apart from the heads, were preserved in ice inside marked plastic bags and the final sample (n=20) was randomly selected.

Age of wild specimens was determined, based on the annual rings of the alcohol embedded otoliths. For this reason, inversed stereoscope with 50X magnification was used and age was determined twice without considering the initial observation. Differential determination of age led to exclude the respective otoliths from the sample. Moreover, otoliths with less clear rings were also excluded. From the eighty fish examined, thirty eight fish was 1+ and 2+ and twenty of them were randomly selected. Accordingly, all procedures were identical to that of farmed specimens.

Moisture content in flesh in both groups of fish was determined by calculating weight deference of a milled body part before and after drying in oven at 103 ± 20 °C for three hours.

Determination of total lipids and fatty acids: Flesh samples were de-frosted, chopped and subsequently homogenized for 1 min in a mechanical homogenizer with metallic blades in low temperature (ice bath). Twenty grams of flesh homogenate and the entire amount of perivisceral homogenate were used from each sample. Lipid extraction was performed by the Bligh & Dyer method [8] using chloroform and methanol in a 2:1 ratio. Subsequently, the homogenized liquid was centrifuged at 3,000 rpm for lipid separation; the solution was removed, weighted and placed in bottle in order to initiate the vacuum evaporation process at 60 °C. The total weight of lipids was estimated from the created sediment using the following formula: Total lipid = weight of lipid aliquot x volume of chloroform layer / volume of aliquot). Subsequently, fatty acids were methyl-esterified with a 12% boron trifluoride methanol solution (BF3-MeOH) (Folch et al., 1957). Methyl-esters were obtained with normal hexane [6]. The analysis was performed using gas chromatography (Model GC-17A; Shimadzu, Kyoto, Japan) with capillary column and ionized flame detector (TRACETM TR-FAME GC Column, Thermo Fisher Scientific Inc.) and automatic sampler (HT 310A, HTA). Pure helium of 82 KPa flow was used for the analysis, air of 50 KPa flow and hydrogen of 60 KPa flow under the following conditions: initial temperature was 150 °C for 5

min, followed by a 5 °C min^{-1} pace until 170 °C for 10 min and then, 5 °C min^{-1} pace until 220 °C for 20 min. The identification of fatty acids (methyl-esters) was performed by comparing the impressed peaks in special PC programme with Qalmix Fish (89-5550) and Methyl Dodecanoate (20-1200) standard fatty acids (Larodan Fine Chemicals AB).

Statistical analysis (mean values, standard deviation and proportions) was performed using Excel 2003 (Microsoft). T-tests were applied after variability comparison by F tests.

3. Results and Discussion

Table 1. Fatty acid profile (% ±SD) of wild and farmed sea bass lipids

Fatty acids	Wild	Farmed
C14:0	2.29±0.25	2.76±0.14
C15:0	0.00±0.00	0.33±0.05
C16:0	20.71±0.97	13.85±0.61
C18:0	7.32±0.75	3.72±0.12
C16:1 n-7 (9C)	5.81±0.86	4.01±0.11
C18:1 n-9 (9C)	18.74±2.44	19.61±0.49
C18:1 n-7 (11C)	4.82±0.57	3.17±0.17
C20:1 n-9 (11C)	0.00±0.00	0.42±0.06
C22:1 n-9 (13C)	9.92±1.13	6.73±0.11
C18:2 n-6	6.39±0.87	18.05±0.09
C18:3 n-3	0.00±0.00	2.09±0.06
C18:4 n-3	0.00±0.00	0.69±0.06
C20:4 n-6	3.64±0.54	0.68±0.04
C20:5 n-3	0.00±0.00	2.15±0.05
C22:4 n-6	0.00±0.00	0.10±0.00
C22:5 n-3	2.80±0.53	1.80±0.05
C22:6 n-3	17.17±0.76	8.62±0.19
Total n-3 fatty acids	19.97±0.64	15.35±0.16
Total n-6 fatty acids	10.03±1.40	18.83±0.12
n-3/n-6 ratio	2.03±0.28	0.82±0.01
EPA/DHA ratio	0.00±0.00	0.25±0.01

Mean moisture values were 74.6 (±1.1%) and 69.1 (±1.8%) in wild and farmed sea bass, respectively. Total fillet lipid content was higher (7.31% ±1.59) for the farmed, compared to the wild fish (1.68% ±1.9), (ANOVA, $P<0.001$). In the same manner, total perivisceral fat was higher in farmed (25.87 ±5.78) compared to wild sea bass (0.11 ±0.02), ANOVA, $P<0.001$.

The analysis of the fillets fatty acid content indicates that wild fish are superior in terms of the nutritional value (Table 1). More specifically, saturated and mono-unsaturated fatty acids (MUFAs) were dominant in farmed sea bass ($P<0.001$) compared to PUFAs in wild specimens. Palmitic acid (C16:0) was the main fatty acid within the saturated fatty acids in both groups of fish, followed by stearic acid (C18:0) and myristic acid (C14:0). The most dominant MUFAs were the oleic acid (C18:1 n-9) followed

by erucic acid (C22:1 n-9), palmitoleic acid (C16:1 n-7) and vaccenic acid (C18:1 n-7) in both wild and farmed fish. Within PUFAs, DHA (C22:6 n-3) was dominant in wild specimens instead of linoleic acid (C18:2 n-6) in farmed specimens, although both fatty acids were present in lower values in both groups. All other fatty acids were identified but in far lower values. Total n-3 fatty acids exceeded n-6 in wild sea bass ($P<0.001$), whereas the opposite was evident in farmed sea bass ($P<0.001$). The n-3/n-6 ratio was in favour of wild sea bass ($P<0.001$) but EPA/DHA ratio was in favour of farmed sea bass. The Ratio EPA/DHA in the flesh of wild sea bass is naught, because non was identification for EPA.

In the perivisceral fat, the dominant fatty acid within MUFAs was C18:1n-9 in both wild and farmed fish, followed by C16:1n-7, C22:1n-9 and C20:1n-9 in wild, and C20:1n-9, C22:1n-9 and C16:1n-7 in farmed fish. PUFAs exceeded in farmed fish ($P<0.001$) and C18:2n-6 was dominant followed by C22:6n-3 in both groups. The n-3 and n-6 were increased ($P<0.001$) in the perivisceral fat of farmed fish. The n-3/n-6 ratio was in favor of the wild sea bass and the EPA/DHA ratio was in favour of the farmed sea bass (Table 2).

Table 2. Fatty acid profile (% ±SD) of total perivisceral fat of wild and farmed sea bass

Fatty acids	Wild N=6	Farmed N=6
C14:0	3.90±0.14	3.00±0.23
C15:0	0.65±0.05	0.38±0.04
C16:0	15.10±0.57	13.00±0.06
C18:0	3.80±0.22	3.52±0.15
C16:1 n-7 (9C)	7.75±0.05	4.18±0.25
C18:1 n-9 (9C)	23.28±1.04	24.13±0.88
C18:1 n-7 (11C)	4.30±0.22	5.48±0.12
C20:1 n-9 (11C)	0.00±0.00	0.00±0.00
C22:1 n-9 (13C)	7.45±0.05	4.40±0.24
C18:2 n-6	9.45±0.16	15.67±0.83
C18:3 n-3	1.55±0.05	1.57±0.12
C18:4 n-3	0.62±0.08	0.73±0.08
C20:4 n-6	2.30±0.33	0.65±0.05
C20:5 n-3	0.00±0.00	4.92±0.31
C22:4 n-6	0.00±0.00	0.17±0.05
C22:5 n-3	2.15±0.16	1.67±0.05
C22:6 n-3	6.35±0.16	5.37±0.26
Total n-3 fatty acids	10.67±0.44	14.25±0.79
Total n-6 fatty acids	11.75±0.49	16.48±0.92
n-3/n-6 ratio	0.91±0.00	0.86±0.01
EPA/DHA ratio	0.00±0.00	0.92±0.02

The fatty acid profile of sea bass fillets and perivisceral fat, indicate the significance of the fatty acid content of aquaculture feeds. The dominance of 18:2n-6, linoleic acid (LA) the increased 18:3 n-3, Linolenic acid (LNA) and the increased n-6/n-3 ratio in the flesh in farmed fish appears to be a result of the inclusion of increased amounts of plant oils as an alternative raw material for fish feeds [4]. As a

result farmed fish exhibited lower nutritional value in terms of FAs, especially in terms of high LA& LNA content and low n-3/n-6 ratio, which is important for human's health. LA and LNA in fish feeds cannot be transformed easily to n-6 and n-3 FAs in the flesh of marine fish, contrary to fresh water fish. This is probably due to lack of necessary enzymes (D-5-6-Desaturase) which elongate the carbonic chain of fatty acids [8]. This disability of marine fish leads to LA and LNA inclusion in flesh lipids, which finally end to the human body through consumption. Studies on the transformation of linoleic acid to AA and of linolenic acid (18:3 n-3) to EPA and DHA in several marine fish species showed that transformation rates were insufficient, precarious and negligible or null [12]. Substitution of natural fish oils with soybean oil showed a remarkable increase of linoleic acid and linolenic acid in the muscle of farmed fish, whereas n-3/n-6 ratio was lower. Moreover, n-3 fatty acids were reduced even by 72 % compared to fish oil feeding fish [5].

The main reason for the use of alternative sources of raw materials in fish feeds is the high cost of fish meal and fish oil due to increasing demand and limited production from wild fish stocks. This trend tends to increase as a convenient tool to reduce production cost in an intensely competitive market and have been reflected in previous published works [4,7] and in FAO reports for decreased imports and consumption of fish meal and fish oil, which during the last decade, decreased by 18% in the European Union, despite the increase of fish farming production. This fact indicates that the alteration of fish feeds composition have been established during the last decade. Nutritional experiments in fish showed that the addition of 60% soybean oil in fish feed resulted in a 428-647% increased levels of LA in the flesh of fish compared to fish oil based feed. In the same manner, 60% linseed oil resulted in increased levels of LA in the flesh of fish compared to controls [6]. Increased LA consumption may cause 100 times greater risk of homicide mortality. Increased LA consumption may contribute to depression and increased cardiac mortality [5].

In several cases, previously reported results of FAs profile for sea bass vary with region, season and feeding regimes [14, 15, 16] the results presented in our study concerning PUFAs profile in wild and farmed specimens alos vary, probably due to differences in the fish samples and in fish feeds consumed, as well as to the rearing and environmental conditions. Nevertheless, the increased presence of MUFAs and especially of oleic acid (C18:1 n-9) and linoleic acid in the flesh of farmed sea bass showed that the fish feed used was rich in plant oils, which are probably harmful both for normal development of fish [10] and consumer's health [11].

The inclusion of plant oils in fish feeds will probably be increased in the course of time as a result of competition and cost reduction efforts in fish farming industry. In this context, strict rules and frequent controls in fish feeds and farmed products will be required to prevent deterioration in product quality and any possible harm to public health.

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4. Conclusions

The results indicate biochemical differences between the wild and farmed sea bass as well between fillet and perivisceral fat. Historically, in comparison to wild, the farmed fish exhibit differences in the fatty acid profile [14,16]. An increased presence of linoleic acid (18:2n-6) and a reduction of n-3/n-6 ratio in farmed sea bass, most likely resulted from fish feeds rich in terrestrial plant oils, which downgraded the nutritional quality of farmed sea bass. The results of the present work indicate that compared to the farmed fish, the wild sea bas exhibits significantly higher concentration of fatty acids with well known health benefits. The nutritional value of wild fish can be improved by exploiting the genetic and dietary induced changes in the fatty acid content of fish [7]. This may require an initial increased cost of research and development as well the increase cost of utilising marine fish oil in the last growing phase of production, but the benefits to the consumers and the marketing of farmed fish could rapidly constitute this economically viable.

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